Culture Handling Procedure

1. When cultures arrive in the laboratory, streak the original culture onto chocolate agar plates and/or Modified Thayer-Martin plates.

2. Incubate the cultures at 35^{o}C, in 5% CO_2, for 24 to 48 hrs.

3. Check for purity. If the culture is pure, subculture on to GCII agar plates and incubate as in step #2. If the culture is not pure, pick 4-5 colonies that appear to be typical of *Neisseria gonorrhoeae*, and again subculture to chocolate and/or Modified Thayer-Martin. Incubate as in step #2.

4. From the pure culture on GCII plates, use a portion of this culture to make a suspension in GC freezing medium.

5. Label cryovials.

6. Dispense the culture suspension into cryovials and freeze at -70^{o}C.

Note: Once cultures are determined to be pure, and added to **your** culture collection, the original culture vials should be discarded.

When using cultures from an already established culture collection:

1. Label all plates or tubes **BEFORE** removing the cultures from the freezer. DO NOT keep cultures out of the freezer for more than one hour.

2. Using a Pasteur pipette, mix the culture then place one drop onto the chocolate agar plate.

3. Return the cultures to the collection, to the exact place from which they were taken.

4. Streak the chocolate agar plate for isolation, then incubate at 35^{o}C, in 5% CO_2, for 18 to 20 hrs.

5. After incubation, inspect cultures for purity.

6. Pick 4-5 well isolated colonies, streak onto GCII. Incubate at 35^{o}C, in 5% CO_2, for 18 to 20 hrs.

7. After incubation, but before proceeding with the other testing procedures, in GC freezing medium, make a suspension of a portion of the culture.

8. Dispense this culture suspension into properly labeled cryovials, freeze at -70^{o}C. This culture vial should be added to **your** collection, and used if repeat isolations are needed.

9. Use the remaining colonies from the GCII plate for testing.